PROCEEDINGS OF THE BIOLOGICAL SOCIETY OF WASHINGTON
108(1):1–5. 1995.

The karyotypes of two South American mouse opossums of the genus *Thylamys* (Marsupialia: Didelphidae), from the Andes, and eastern Paraguay

R. Eduardo Palma

Department of Biology and Museum of Southwestern Biology, University of New Mexico, Albuquerque, New Mexico 87131-1091, U.S.A.

Abstract.—The karyotypes of *Thylamys macrura,* from eastern Paraguay and adjacent Brazil, and *T. pallidior,* from the Andean Altiplano in Argentina and Bolivia, are reported for the first time. The karyotypes of somatic tissues from female *T. macrura* and female *T. pallidior* consist of 2N = 14. However, the diploid number found in somatic cells in males of *T. pallidior* exhibits 2N = 13 because the Y chromosome is missing. The missing Y phenomenon might represent another case of chromosome mosaicism in American marsupials, although this time detected in didelphids.

Tate (1933) recognized five species groups in the genus *Marmosa* Gray, 1821 (sensu lato) based on phenetic characters. Subsequent morphologic, chromosomic, and serologic studies have shown that these assemblages approximate genera (Reig et al. 1985, 1987; Gardner & Creighton 1989). Among these, *Thylamys* Gray, 1843, has the southernmost distribution of mouse opossums in South America encompassing elevations as high as 3500 m, as well as lower temperate and subtropical habitats (Cabrera 1958). *Thylamys* differs from other marmosines in having a characteristic tricolored dorsal pattern, and the capacity to store fat in the tail (Tate 1933, Mann 1978) Recent revisions of the genus recognize five species (Gardner & Creighton 1989, Gardner 1993): *Thylamys pusillus* (Desmarest 1804), *Thylamys macrura* (Olfers 1818), *Thylamys elegans* (Waterhouse 1839), *Thylamys velutinus* (Wagner 1842), and *Thylamys pallidior* (Thomas 1902). *Thylamys pallidior* occurs on the rocky slopes of the Altiplano of Bolivia and Argentina (Tate 1933; fig. 1), whereas, the subtropical form *T. macrura* is found in the humid forests of eastern Paraguay and adjacent Brazil (Gardner 1993; fig. 1). This constitutes the sixth published record of this species for Paraguay, since the original description by Olfers (1818) based on Azara's (1801) "Micouré à queue longue," or "Colilargo" (Azara 1845). Tate (1933) referred to this species as *Marmosa marmota,* and Cabrera (1958) as *Marmosa grisea.* The name *macrura* (Olfers 1818) is available and *T. macrura* is used by Gardner (1993). In this study, the karyotypes of *T. macrura* and *T. pallidior* are reported for the first time. This is the first study documenting the absence of one of the sex chromosomes of somatic cells in didelphid marsupials.

Methods

Chromosomal preparations were obtained directly from bone marrow using the standard velban technique described in Anderson et al. (1987). A minimum of 10 metaphase spreads were counted for each specimen. Nomenclature for chromosome morphology and fundamental number (FN) follows Patton (1967). Five specimens of *T. pallidior* from two localities in Bolivia were analyzed. The karyotype of *T. macrura* was

Fig. 1. Map showing the distribution of *T. pallidior* along the Andes of Argentina and Bolivia, and *T. macrura* in eastern Paraguay. The boxes and dot represent the collecting site given in the text.

obtained from a specimen from eastern Paraguay. Voucher specimens, chromosome slides, and cell suspensions are deposited in the Museum of Southwestern Biology (MSB) and the American Museum of Natural History (AMNH). Collection localities are (Fig. 1): Bolivia: Department of Chuquisaca, Camargo, 68 km (by road) N of Camargo, 3400 m, 20°09′S, 65°17′W (3 males MSB 57003, AMNH 262406, and AMNH 262407; 1 female AMNH 262405); Department of Tarija, Serranía Sama, 3200 m, 21°27′S, 64°52′W (1 female, AMNH 263555). Paraguay: Department of Concepción, Escuela Agropecuaria, 7 km (by road) NE from Concepción, 23°21′S, 57°23′W (1 female, MSB NK 27536).

Results and Discussion

The autosomes of *T. macrura* (2N = 14, FN = 20; Fig. 2a) consist of three pairs of large submetacentrics (1–3), one pair of medium-sized metacentrics (4), and two pairs of small acrocentrics (5–6). The X chromosome is a small acrocentric. The autosomes of *T. pallidior* (Fig. 2b) are not distinguishable from those of *T. macrura,* although the three males of the Andean species present 2N = 13, FN = 20; the Y chromosome was absent in all counted plates. The female *T. pallidior* exhibited the complete set of chromosomes, 2N = 14. A male of *T. macrura* was not available for karyotype.

The autosomic complement of the species of *Thylamys* reported here is similar to those documented previously for other species of the genus, such as *T. elegans* from Chile and Bolivia, which possess the identical three group pattern of autosomes and morphology (Reig et al. 1972, Palma & Yates 1995). *Micoureus cinereus* and *M. constantiae* have similar diploid and fundamental number (2N = 14, FN = 20; Palma & Yates 1995). *Marmosa* (sensu stricto), *Marmosops,* and *Gracilinanus,* have 2N = 14, however the fundamental number in these taxa is FN = 24 (Reig 1968, Palma & Yates 1995).

The mouse opossum karyotypes presented in this paper reinforce the concept of chromosomic conservatism in marsupials, and support the fact that marsupial species that occur in remarkably different habitats share a common karyotype (Reig et al. 1977, Hayman 1990). The common 2N = 14 is shared by most of the marmosines in the Neotropics. *Marmosa canescens* (2N = 22), is the only known exception (Engstrom & Gardner 1988).

The conclusive evidence of the absence of the Y chromosome in *T. pallidior* is difficult to determine under the methodology followed in this study. It is possible that the Y has been translocated to another chromosome, or this condition may be another example of chromosome mosaicism, i.e., a difference in sex-chromosome presence between the germ line and cells of the somatic tissues (Hayman 1990). Similar patterns of absence of the Y chromosome for somatic

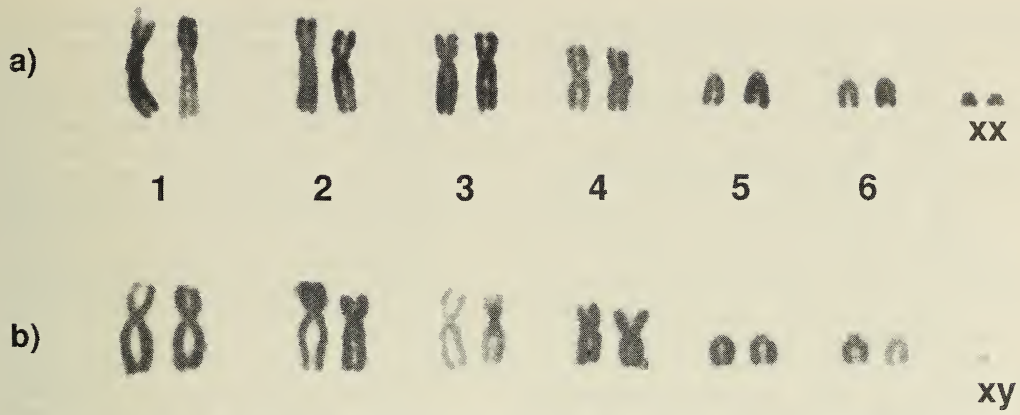

Fig. 2. a, Standard karyotype of a female *Thylamys macrura* from Concepción, Paraguay, 2N = 14, FN = 20. b, Standard karyotype of a male *Thylamys pallidior* from Camargo, Chuquisaca, Bolivia, 2N = 14, FN = 20.

cells have been found for *Chironectes minimus* (Palma & Yates 1995) and *Dromiciops australis* (Gallardo & Patterson 1987). Additional reports of chromosome mosaicism have been also made for Australian marsupials of the family Peramelidae, where one of the X chromosomes is missing in somatic cells of females, and in *Petauroides* (Petauridae), where the Y chromosome is missing from the majority of cells obtained from bone marrow (Murray et al. 1979, Hayman 1990).

The missing Y chromosome found in *Dromiciops australis* (Gallardo & Patterson 1987), caused these authors to suggest that the microbiotheriid would be more related to Australasian marsupials than to American metatherians, supporting Szalay's (1982) contention that *Dromiciops* and Australian marsupials constitute the cohort Australidelphia. Data from this study and from Palma & Yates (1995), prove that the Y chromosome is missing not only in *Dromiciops* and Australasian marsupials, but in American marsupials as well. This scenario fits a typical case of parallelism or represents a plesiomorphic condition in the evolution of metatherian sexual chromosomes of both geographic regions. Hence, this character cannot be used as evidence for inferring phylogeny between Australian and American marsupial lineages.

Acknowledgments

Mike Bogan, Jennifer K. Frey, Jorge Salazar, and Terry L. Yates provided valuable comments on a draft of the manuscript. Special thanks to Lucy Aquino from CITES-Paraguay, and Carl Shuster; to Flaviano Colman and Luis Morán of the National Museum of Natural History of Paraguay, Maribé Robles, and to the National Parks Office of the Ministry of Agriculture and Livestock of Paraguay. Funding support was provided by a NSF Research Improvement Grant PDS-105-774, the Latin American Institute, University of New Mexico, and Sigma Xi. Field work in Bolivia was supported by NSF grants BSR-83-16740 to the American Museum of Natural History (S. Anderson), and BSR-84-08923 to the University of New Mexico (T. L. Yates). The map was prepared by Beth Dennis.

Literature Cited

Anderson, S., T. L. Yates, & J. A. Cook. 1987. Notes of Bolivian mammals 4: the genus *Ctenomys* (Rodentia, Ctenomyidae) in the eastern lowlands.—American Museum Novitates 2891:1–20.

Azara, F., d'. 1801. Essais sur l'histoire naturelle des quadrupèdes de la Province du Paraguay. Traduits sur le manuscript inédit de l'auteur, Pra. M. L. E. Moreau-Saint-Méry. Charles Pougens, Paris, 366 pp.

———. 1845. Viajes por la América del Sur. Biblioteca de Impresos Raros Americanos. Universidad de la República, Facultad de Humanidades y Ciencias, Instituto de Investigaciones Históricas, Montevideo 1982, 318 pp.

Cabrera, A. 1958. Catálogo de los mamíferos de América del Sur.—Revista del Museo Argentino de Ciencias Naturales "Bernardino Rivadavia", Ciencias Zoológicas 4:1–307.

Desmarest, A. G. 1804. Taubleau méthodique des mammifères. Pp. 5–38 *in* Nouveau dictionnaire d'histoire naturelle, appliquée aux arts, principalement à l'agriculture, à l'économie rurale et domestique: Par une société de naturalistes et d'agriculteurs: Avec des figures tirées des trois règnes de la nature. Vol. 24. Paris, Deterville.

Engstrom, M. D., & A. L. Gardner. 1988. Karyotype of *Marmosa canescens* (Marsupialia: Didelphidae): a mouse opossum with 22 chromosomes.—The Southwestern Naturalist 33:230–233.

Gallardo, M. H., & B. D. Patterson. 1987. An additional 14-chromosome mosaicism in South American marsupials. Pp. 111–115 *in* B. D. Patterson & R. M. Timm, eds., Studies in Neotropical mammalogy: essays in honor of Philip Hershkovitz.—Fieldiana: Zoology, new series 39:1–506.

Gardner, A. L. 1993. Order Didelphimorphia. Pp. 15–23 *in* D. E. Wilson & D. M. Reeder, eds., Mammal species of the world: a taxonomic and geographic reference. Second edition. Smithsonian Institution Press, Washington, 1206 pp.

———, & G. K. Creighton. 1989. A new generic name for Tate's (1933) *microtarsus* group of South American mouse opossums (Marsupialia: Didelphidae).—Proceedings of the Biological Society of Washington 102:3–7.

Gray, J. E. 1821. On the natural arrangement of the vertebrose animals.—London Medical Repository 15: 296–310.

———. 1843. List of the specimens of mammalia in the collection of the British Museum of London. British Museum (Natural History), 216 pp.

Hayman, D. L. 1990. Marsupial cytogenetics.—Australian Journal of Zoology 37:331–349.

Mann, G. 1978. Los pequeños mamíferos de Chile.—Gayana, Zoología 40:1–342.

Murray, J. D., G. M. Mckay, & G. B. Sharman. 1979. Studies on metatherian sex chromosomes. IX. Sex chromosomes of the greater glider (Marsupialia: Petauridae).—Australian Journal of Biological Sciences 32:375–386.

Olfers, I. 1818. Bemerkungen zu Illiger's Ueberblick der Säugthiere, nach ihrer Vertheilung über die Welttheile, rücksichtlich der Südamerikanischen Arten (Species). Pp. 129–137 *in* Journal von Brazilien, oder vermischte Nachrichten aus Brasilien, auf wissenschaftlichen Reisen gesammelt, W. L. Eschwege, 129–137 *in* F. J. Bertuch, ed., Neue Bibliotek der wichtigsten Reisebeschreibungen zur Erweiterung der erd- und Völkerkunde; in Verbindung mit einigen anderen Gelehrten gesammelt und herausgegeben. Weimar: Verlage des Landes-Industrie-Comptoirs. Vol. 15, 304 pp.

Palma, R. E., & T. L. Yates. 1995. The chromosomes of Bolivian didelphid marsupials.—Occasional Papers, The Museum, Texas Tech University, Lubbock (in press).

Patton, J. L. 1967. Chromosome studies of certain pocket mice, genus *Perognathus* (Rodentia, Heteromyidae).—Journal of Mammalogy 48:27–37.

Reig, O. A. 1968. The chromosomes of the didelphiid *Marmosa robinsoni* Bangs.—Experientia 24: 185–186.

———, R. Fernández, & A. Spotorno. 1972. Further occurrence of a karyotype of 2n = 14 chromosome in two species of Chilean didelphoid marsupials.—Zeitschrift für Säugetierkunde 37:37–42.

———, J. A. W. Kirsch, & L. Marshall. 1985. New conclusions on the relationships of the opossum-like marsupials with an annotated classification of the Didelphimorphia.—Ameghiniana 21:335–343.

———, ———, & ———. 1987. Systematic relationships of the living and Neocenozoic American "opossum-like" marsupials (suborder Didelphimorphia), with comments on the classification of these and of the Cretaceous and Paleogene New World and European metatherians. Pp. 1–89 *in* M. Archer, ed., Possums and opossums: studies in evolution. Surrey Beatty and Sons and the Royal Society of New South Wales, Sydney, 1 and 2, 800 pp.

———, A. L. Gardner, N. O. Bianchi, & J. L. Patton. 1977. The chromosomes of the Didelphidae (Marsupialia) and their evolutionary significance.—Biological Journal of the Linnean Society 9:191–216.

Rofe, R., & D. Hayman. 1985. G-banding evidence for a conserved complement in the Marsupialia.—Cytogenetics and Cell Genetics 39:40–50.

Szalay, F. S. 1982. A new appraisal of marsupial phylogeny and classification. Pp. 621–640 *in* M. Archer, ed., Carnivorous marsupials. The Royal Biological Society of New South Wales, Sydney, Australia, 802 pp.

Tate, G. H. H. 1933. Systematic revision of the marsupial genus *Marmosa*, with a discussion of the

adaptive radiation of the murine opossums (*Marmosa*).—Bulletin of the American Museum of Natural History 66:1–250.

Thomas, O. 1902. On *Marmosa marmota* and *elegans* with descriptions of new subspecies of the latter.—Annals and Magazine of Natural History, series 7, 9:408–410.

Wagner, J. A. 1842. Diagnosen neuer Arten brasilischer Säugethiere.—Archiv Naturgesch 8:356–362.

Waterhouse, G. R. 1839. Mammalia. Pp. 49–97 *in* C. Darwin, ed., The zoology of the voyage of the H. M. S. Beagle under the command of Captain Fitzroy, R. N., during the years 1832–1836. Smith, Elder and Company, London, 97 pp.